

<u>DB Name</u>	<u>Query</u>	<u>Hit Count</u>	<u>Set Name</u>
USPT,JPAB,EPAB,DWPI	16 near10 (attach\$8)	6	<u>L14</u>
USPT,JPAB,EPAB,DWPI	16 same (adapter\$1 or linker\$1 or oligonucleotide\$1 or primer\$1)	5	<u>L13</u>
USPT,JPAB,EPAB,DWPI	16 near10 (adapter\$1 or linker\$1 or oligonucleotide\$1 or primer\$1)	1	<u>L12</u>
USPT,JPAB,EPAB,DWPI	16 near10 (adapter\$1 or linker\$1 or oligonucleotide\$1)	1	<u>L11</u>
USPT,JPAB,EPAB,DWPI	15 same 12	0	<u>L10</u>
USPT,JPAB,EPAB,DWPI	18 not 15	2	<u>L9</u>
USPT,JPAB,EPAB,DWPI	13 same 16	4	<u>L8</u>
USPT,JPAB,EPAB,DWPI	13 near10 16	0	<u>L7</u>
USPT,JPAB,EPAB,DWPI	14 near5 site\$1	97	<u>L6</u>
USPT,JPAB,EPAB,DWPI	14 near10 13	4	<u>L5</u>
USPT,JPAB,EPAB,DWPI	site adj specific adj recombination	375	<u>L4</u>
USPT,JPAB,EPAB,DWPI	11 or 12	24617	<u>L3</u>
USPT,JPAB,EPAB,DWPI	(nucleic adj acid\$1) near10 amplif\$8	7287	<u>L2</u>
USPT,JPAB,EPAB,DWPI	PCR or (polyerase adj chain adj reaction\$1)	22358	<u>L1</u>

**WEST**

Generate Collection

L5: Entry 3 of 4

File: USPT

Nov 23, 1999

US-PAT-NO: 5989872

DOCUMENT-IDENTIFIER: US 5989872 A

TITLE: Methods and compositions for transferring DNA sequence information among vectors

DATE-ISSUED: November 23, 1999

US-CL-CURRENT: 435/91.2; 435/325, 435/354, 435/366, 435/455, 435/463, 435/465,  
435/91.4, 435/91.41, 536/23.1, 536/24.1, 536/24.2, 536/24.33

APPL-NO: 8/ 909525

DATE FILED: August 12, 1997

**WEST**☐ **Generate Collection**

L5: Entry 3 of 4

File: USPT

Nov 23, 1999

US-PAT-NO: 5989872

DOCUMENT-IDENTIFIER: US 5989872 A

TITLE: Methods and compositions for transferring DNA sequence information among vectors

DATE-ISSUED: November 23, 1999

## INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Luo; Ying	Los Altos	CA	N/A	N/A
Hua; Shaobing	Cupertino	CA	N/A	N/A
Zhu; Li	Palo Alto	CA	N/A	N/A

US-CL-CURRENT: 435/91.2; 435/325, 435/354, 435/366, 435/455, 435/463, 435/465,  
435/91.4, 435/91.41, 536/23.1, 536/24.1, 536/24.2, 536/24.33

## CLAIMS:

What is claimed is:

1. A method of transferring DNA sequence information from a first vector to a second vector, said method comprising:  
contacting said first vector with a set of three pairs of oligonucleotide primers under conditions sufficient to produce three different PCR products, wherein each oligonucleotide primer comprises a first region of sequence identity with said first vector and a second region which does not hybridize with said first vector and provides for homologous recombination with said second vector, whereby three different PCR products are produced; and  
contacting said three different PCR products with said second vector under conditions sufficient for homologous recombination to occur;  
whereby said DNA sequence information is transferred from said first vector to said second vector.
2. The method according to claim 1, wherein said DNA sequence information is the sequence of an EST clone.
3. The method according to claim 1, wherein said three different PCR products correspond to three different reading frames.
4. The method according to claim 1, wherein each pair of said set of oligonucleotide primers is contacted with said first vector at substantially the same time.
5. The method according to claim 1, wherein each pair of said set of oligonucleotide primers is contacted with said first vector at different times.
6. A second vector produced according to claim 1, wherein each of the first regions in said set of primers is the same length.
7. A method of transferring DNA sequence information from a first plasmid to a second plasmid, said method comprising:  
contacting a first plasmid with a set of three pairs of oligonucleotide primers under conditions sufficient to produce three different PCR products, wherein each PCR product corresponds to a different reading frame and each oligonucleotide primer comprises a first region of sequence identity with said first plasmid and a second region which does not hybridize with said first plasmid and provides for homologous recombination with said second plasmid, whereby three different PCR products are produced; and  
contacting said three different PCR products, with said second plasmid under conditions sufficient for homologous recombination to occur;  
whereby said DNA information is transferred from said first plasmid to said second plasmid.

8. The method according to claim 7, wherein each pair of said set of oligonucleotide primers is contacted with said first plasmid at substantially the same time.
9. The method according to claim 7, wherein each pair of said set of oligonucleotide primers is contacted with said first plasmid at different times.
10. The method according to claim 7, wherein said second plasmid is a DNA sequence encoding either a DNA binding domain or an activation domain of a eukaryotic transcriptional activator.
11. A second plasmid produced according to claim 7, wherein each of the first regions in said set of primers is the same length.
12. A method of transferring DNA sequence information from a first plasmid into an expression plasmid, said method comprising:  
contacting said first plasmid with a set of three pairs of oligonucleotide primers under conditions sufficient to produce three different PCR products, wherein each PCR product corresponds to a different reading frame and each oligonucleotide primer comprises a first region of sequence identity with said first plasmid and a second region which does not hybridize with said first plasmid and provides for homologous recombination with said expression plasmid, whereby three different PCR products are produced; and  
co-transforming said three different PCR products and said expression plasmid into a yeast host whereby homologous recombination occurs, wherein said expression plasmid comprises a DNA sequence encoding either a DNA binding domain or an activation domain of a eukaryotic transcriptional activator;  
whereby said DNA sequence information is transferred into said expression plasmid.
13. The method according to claim 12, wherein each pair of said set of oligonucleotide primers is contacted with said first plasmid at substantially the same time.
14. The method according to claim 12, wherein each pair of said set of oligonucleotide primers is contacted with said first plasmid at different times.
15. The method according to claim 12, wherein said eukaryotic transcriptional activator is GAL4.
16. The method according to claim 12, wherein said second region ranges in length from 20 to 80 bp.
17. An expression plasmid produced according to claim 12, wherein each of the first regions in said set of primers is the same length.
18. The method according to claim 16, wherein said second region does not exceed 50 bp in length.
19. A kit for use in transferring DNA sequence information from a first vector to a second vector, said kit comprising:  
a set of three pairs of oligonucleotide primers, wherein each primer comprises a first region of sequence identity with said first vector and a second region which does not hybridize with said first vector and provides for homologous recombination with said second vector.
20. The kit according to claim 19, wherein said second vector is an expression plasmid comprising a sequence encoding either a DNA binding domain or an activation domain of a eukaryotic transcriptional activator.
21. The kit according to claim 20, wherein said kit further comprises two different expression plasmids, wherein each plasmid comprises said sequence encoding either said DNA binding domain or said activation domain.
22. A kit for use in transferring DNA sequence information from a first plasmid to an expression plasmid, said kit comprising:  
an expression plasmid comprising a sequence encoding either a GAL4 DNA binding domain or a GAL4 activation domain;  
a set of three pairs of oligonucleotide primers, wherein each primer comprises a first region of sequence identity with a first plasmid and a second region which does not hybridize with said first plasmid and provides for homologous recombination with said expression plasmid.

Transformation genetique; Inversion; Deletion; Translocation;  
Rearrangement genetique; Article synthese

Classification Codes: 002A31C02A9; 215

? ds

Set	Items	Description
S1	2958	(SITE (3N) SPECIFIC) (5N) RECOMBINATION
S2	47541	INTEGRASE? OR CRE OR INT OR FLP OR RESOLVASE? OR INTEGRON?
		OR RECOMBINASE?
S3	29243	(RECOGNITION (5N) (SITE? OR SEQUENCE? OR ADAPTER? OR LINKE- R?))
S4	1454	S1 AND S2
S5	90	S4 AND S3
S6	0	S5 AND DT=REVIEW
S7	1	S5 AND REVIEW
S8	1	PCR AND S5
S9	67	S5 AND PY<1997
S10	6	S9 AND CLON?
S11	493	(ATTACH? OR FLANK? OR PLACING OR PUTTING) (10N) S3
S12	402	S11 AND PY<1998
S13	0	12 DR
S14	237	REMOVE DUPLICATES S12 (unique items)
S15	6	S14 AND PCR
S16	201	S2 (5N) S3
S17	2	S16 (5N) PCR
S18	120	S16 NOT (VECTOR? OR PLASMID?)
S19	98	S18 AND PY<1998
S20	60	REMOVE DUPLICATES S19 (unique items)
S21	0	S20 AND PCR
S22	1	S20 AND (PRIMER OR PRIMERS)
S23	8	S20 AND OLIGONUCLEOTIDE?
S24	52	S20 NOT S23
S25	15	S1 AND S24
		?

A new lambda RES vector with a built-in Tn1721-encoded excision system.

Altenbuchner J

Institut für Industrielle Genetik, Universität Stuttgart, Germany.

Gene (NETHERLANDS) Jan 15 1993, 123 (1) p63-8, ISSN 0378-1119

Journal Code: FOP

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9304

Subfile: INDEX MEDICUS

A new lambda replacement vector for construction of genomic libraries was developed which allows the excision of **cloned** fragments by **site-specific recombination** from the lambda DNA and conversion into autonomously replicating plasmids. The vector system, derived from lambda EMBL4, is called lambda RES. It contains two **recognition** sites for **site-specific recombination** from Tn1721 on both sides of the replacement fragment of lambda EMBL4. Additionally, on one side, there is a plasmid replication origin from Rts1 with a kanamycin-resistance (KmR) marker. DNA fragments in the range of 8-14 kb may be inserted between BamHI or Sall sites in the lambda vector. Efficient excision and conversion of plaque-forming units into KmR colonies are obtained by infection of Escherichia coli strains harbouring Tn1739tnpR on a F' plasmid. Tn1739tnpR is a derivative of Tn1721 with a chloramphenicol-resistance-encoding gene (CmR), the lambda cI repressor gene, and a further copy of the **resolvase-enco**

Mobile gene cassettes and **integrans**: Capture and spread of genes by **site-specific recombination**.

AUTHOR: Hall Ruth M(a); Collis Christina M

AUTHOR ADDRESS: (a)CSIRO Div. Biomol. Eng., Sydney Lab., PO Box 184, North Ryde 2113, NSW\*\*Australia

JOURNAL: Molecular Microbiology 15 (4):p593-600 1995

ISSN: 0950-382X

DOCUMENT TYPE: Literature Review

RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: An **integron** is a genetic unit that includes the determinants of the components of a **site-specific recombination** system capable of capturing and mobilizing genes that are contained in mobile elements called gene cassettes. An **integron** also provides a promoter for expression of the cassette genes, and **integrans** thus act both as natural **cloning** systems and as expression vectors. The essential components of an **integron** are an **int** gene encoding a site-specific **recombinase** belonging to the **integrase** family, an adjacent site, attI, that is recognized by the **integrase** and is the receptor site for the cassettes, and a promoter suitably oriented for expression of the cassette-encoded genes. The cassettes are mobile elements that include a gene (most commonly an antibiotic-resistance gene) and an **integrase-specific recombination site** that is a member of a family of sites known as 59-base elements. Cassettes can exist either free in a circularized form or integrated at the attI site, and only when integrated is a cassette formally part of an **integron**. A single **site-specific recombination** event involving the **integron-associated attI site** and a cassette-associated 59-base element leads to insertion of a free circular cassette into a recipient **integron**. Multiple cassette insertions can occur, and **integrans** containing several cassettes have been found in the wild. The **integrase** also catalyses excisive recombination events that can lead to loss of cassettes from an **integron** and generate free circular cassettes. Due to their ability to acquire new genes, **integrans** have a clear role in the evolution of the genomes of the plasmids and transposons that contain them. However, a more general role in evolution is also likely. Events involving **recombination** between a **specific 59-base-element site** and a nonspecific secondary site have recently been shown to occur. Such events should lead either to the insertion of cassettes at non-specific sites or to the formation of stable cointegrates between different plasmid molecules, and a cassette situated outside the **integron** context has recently been identified.

DESCRIPTORS:

MAJOR CONCEPTS: Enzymology (Biochemistry and Molecular Biophysics);

Evolution and Adaptation; Genetics; Molecular Genetics (Biochemistry and Molecular Biophysics); Pharmacology; Physiology

BIOSYSTEMATIC NAMES: Bacteria-General Unspecified--Eubacteria, Bacteria

ORGANISMS: bacteria (Bacteria - General Unspecified)

BIOSYSTEMATIC CLASSIFICATION (SUPER TAXA): bacteria; eubacteria; microorganisms

MISCELLANEOUS TERMS: ANTIBIOTIC RESISTANCE GENES; CASSETTE ENCODED GENES; EVOLUTIONARY FUNCTION; **INT** GENE; NONSPECIFIC CASSETTE INSERTION; PLASMID; PROMOTER; **RECOGNITION ATT I SITE**; **RECOMBINASE**; TRANSPOSON

CONCEPT CODES:

01500 Evolution

10300	Replication, Transcription, Translation
10806	Enzymes-Chemical and Physical
31000	Physiology and Biochemistry of Bacteria
31500	Genetics of Bacteria and Viruses
38504	Chemotherapy-Antibacterial Agents
10062	Biochemical Studies-Nucleic Acids, Purines and Pyrimidines
10506	Biophysics-Molecular Properties and Macromolecules
12512	Pathology, General and Miscellaneous-Therapy (1971- )
22002	Pharmacology-General
36002	Medical and Clinical Microbiology-Bacteriology

BIOSYSTEMATIC CODES:

05000	Bacteria-General Unspecified (1992- )
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25/9/4 (Item 4 from file: 155)  
DIALOG(R) File 155:MEDLINE(R)  
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08582448 95388520

Chemical probe and missing nucleoside analysis of Flp recombinase bound to the recombination target sequence.

Kimball AS; Kimball ML; Jayaram M; Tullius TD  
Department of Biology, Johns Hopkins University, Baltimore, MD 21218, USA.

Nucleic acids research (ENGLAND) Aug 11 1995, 23 (15) p3009-17,  
ISSN 0305-1048 Journal Code: O8L

Contract/Grant No.: GM 41930, GM, NIGMS

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9512

Subfile: INDEX MEDICUS

The Flp protein catalyzes a site-specific recombination reaction between two 47 bp DNA sites without the assistance of any other protein or cofactor. The Flp recognition target (FRT) site consists of three nearly identical sequences, two of which are separated by an 8 bp spacer sequence. In order to gain insight into this remarkable protein-DNA interaction we used a variety of chemical probe methods and the missing nucleoside experiment to examine Flp binding. Hydroxyl radical footprints of Flp bound to a recombinationally-competent site fall on opposite faces of canonical B-DNA. The 8 bp spacer region between the two Flp binding sites becomes reactive towards 5-phenyl-1,10-phenanthroline.copper upon Flp binding, indicating that once bound by Flp, this segment of DNA is not in the B-form. Missing nucleoside analysis reveals that within each binding site the presence of two nucleosides on the top strand and four on the bottom, are required for formation of a fully-occupied FRT site. In contrast, loss of any nucleoside in the three binding sites in the FRT interferes with formation of lower-occupancy complexes. DNA molecules with gaps in the 8 bp spacer region are over-represented in complexes with either two or three binding sites occupied by Flp, evidence that DNA flexibility facilitates the cooperative interaction of Flp protomers bound to a recombinationally-active site.

Tags: Support, Non-U.S. Gov't; Support, U.S. Gov't, Non-P.H.S.; Support, U.S. Gov't, P.H.S.

Descriptors: \*DNA--Metabolism--ME; \*DNA Nucleotidyltransferases  
--Metabolism--ME; \*DNA-Binding Proteins--Metabolism--ME; \*Recombination,  
Genetic; Base Sequence; Binding Sites; Copper; DNA--Chemistry--CH; DNA  
Nucleotidyltransferases--Genetics--GE; DNA-Binding Proteins--Genetics--GE;  
Edetic Acid--Analog and Derivatives--AA; Hydroxyl Radical; Iron Chelating  
Agents; Molecular Sequence Data; Nucleosides; Phenanthrolines; Recombinant  
Proteins--Biosynthesis--BI

CAS Registry No.: 0 (methidiumpropyl-EDTA-iron(II)); 0 (DNA-Binding  
Proteins); 0 (Iron Chelating Agents); 0 (Nucleosides); 0  
(Phenanthrolines); 0 (Recombinant Proteins); 3352-57-6 (Hydroxyl  
Radical); 60-00-4 (Edetic Acid); 6153-89-5 (5-phenyl-1,10-phenanthroline  
); 7440-50-8 (Copper); 7758-98-7 (Copper Sulfate); 9007-49-2 (DNA)  
Enzyme No.: EC 2.7.7.- (DNA Nucleotidyltransferases); EC 2.7.7.- (FLP  
recombinase)

25/9/7 (Item 7 from file: 155)  
DIALOG(R) File 155:MEDLINE(R)  
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08037405 95034697

Use of mutated FLP recognition target (FRT) sites for the exchange of expression cassettes at defined chromosomal loci.

Schlake T; Bode J

GBF, Gesellschaft für Biotechnologische Forschung mbH, Braunschweig-Stockheim, Germany.

Biochemistry (UNITED STATES) Nov 1 1994, 33 (43) p12746-51, ISSN 0006-2960 Journal Code: A0G

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9502

Subfile: INDEX MEDICUS

Using the FLP/FRT system for site-specific recombination and the wild-type recognition site (FRT) in conjunction with certain mutant FRT sites, it becomes possible to provoke, with high yield, a double-reciprocal crossover event in cultured mammalian cells. It is demonstrated that this technology enables a targeting of expression cassettes to appropriate chromosomal reference sites in the recipient cell to improve the concepts of reverse genetics. The design of mutant FRT sites promoting such a process will be delineated. Our results show that the five spacer mutations tested are functional as the wild type but differ in the extent of their cross-recombination, which has to be minimized for their simultaneous usage.

Tags: Animal

Descriptors: \*DNA--Chemistry--CH; \*DNA Nucleotidyltransferases --Metabolism--ME; \*Mutation; beta-Galactosidase--Genetics--GE; Base Sequence; Binding Sites; Cell Line; DNA--Genetics--GE; DNA--Metabolism--ME; Hamsters; Kidney; Molecular Sequence Data; Mutagenesis; Polymerase Chain Reaction; Recombination, Genetic; Repetitive Sequences, Nucleic Acid; Transfection; Transferases

CAS Registry No.: 9007-49-2 (DNA)

Enzyme No.: EC 2. (Transferases); EC 2.7.7.- (DNA Nucleotidyltransferases); EC 2.7.7.- (FLP recombinase); EC 3.2.1.23 (beta-Galactosidase)

25/9/8 (Item 8 from file: 155)

DIALOG(R) File 155:MEDLINE(R)

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07844801 94111994

Specificity of DNA recognition in the nucleoprotein complex for site-specific recombination by Tn21 resolvase.

Hall SC; Halford SE

Department of Biochemistry, University of Bristol, UK.

Nucleic acids research (ENGLAND) Dec 11 1993, 21 (24) p5712-9, ISSN 0305-1048 Journal Code: O8L

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 9404

Subfile: INDEX MEDICUS

Resolvases from Tn3-like transposons catalyse site-specific recombination at res sites. Each res site has 3 binding sites for resolvase, I, II, and III. The res sites in Tn3 and Tn21 have similar structures at I and II but they differ at III. Mutagenesis of the Tn21 res site showed that sub-site III is essential for recombination though the sequences in III that are recognized by Tn21 resolvase are positioned differently from the equivalent sequences in the Tn3 site. The deletion of III caused a 1,000-fold drop in the rate of recombination. But other mutations at III, changing 3 or 4 consecutive base pairs, caused only 1.5- to 4-fold decreases in rate, even when the mutations were in target sequences for this helix-turn-helix protein. The reason why Tn21 resolvase has similar activities at a number of different DNA sequences may be due to the multiplicity of protein-protein and protein-DNA interactions in its

recombinogenic complex. This lack of precision may be a general feature of nucleoprotein complexes.

Tags: Support, Non-U.S. Gov't

Descriptors: \*DNA--Metabolism--ME; \*DNA Transposable Elements; \*Nucleoproteins--Metabolism--ME; \*Nucleotidyltransferases--Metabolism--ME; Base Sequence; Molecular Sequence Data; Mutagenesis, Insertional; Recombination, Genetic

CAS Registry No.: 0 (DNA Transposable Elements); 0 (Nucleoproteins); 9007-49-2 (DNA)

Enzyme No.: EC 2.7.7 (Nucleotidyltransferases); EC 2.7.7.- (Transposase)

25/9/12 (Item 12 from file: 155)

DIALOG(R)File 155:MEDLINE(R)

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04157626 85153057

The bacteriophage P1 site-specific recombinase cin: recombination events and DNA recognition sequences.

Iida S; Huber H; Hiestand-Nauer R; Meyer J; Bickle TA; Arber W

Cold Spring Harbor symposia on quantitative biology (UNITED STATES) 1984, 49 p769-77, ISSN 0091-7451 Journal Code: DMT

Languages: ENGLISH

Document type: JOURNAL ARTICLE

JOURNAL ANNOUNCEMENT: 8507

Subfile: INDEX MEDICUS

Tags: Support, Non-U.S. Gov't

Descriptors: \*Coliphages--Enzymology--EN; \*DNA Nucleotidyltransferases--Genetics--GE; \*DNA, Viral--Genetics--GE; \*Genes, Viral; \*Recombination, Genetic; Base Sequence; Binding Sites; Coliphages--Genetics--GE

CAS Registry No.: 0 (DNA, Viral)

Enzyme No.: EC 2.7.7.- (cin recombinase); EC 2.7.7.- (DNA Nucleotidyltransferases)

25/9/15 (Item 1 from file: 144)

DIALOG(R)File 144:Pascal

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12134363 PASCAL No.: 95-0366568

Genome manipulation through site-specific recombination

OW D W; MEDBERRY S L

USDA, plant gene expression cent., Albany CA 94710, USA

Journal: Critical reviews in plant sciences, 1995, 14 (3) 239-261

ISSN: 0735-2689 CODEN: CRPSD3 Availability: INIST-20941;

354000051011270030

No. of Refs.: 2 p.1/2

Document Type: P (Serial) ; A (Analytic)

Country of Publication: USA

Language: English

Several DNA site-specific recombination systems have been shown to function in higher eukaryotic cells. These two-component systems consist of a single-polypeptide recombinase and a short recognition sequence of less than 35 bp. Strategic placement of the recognition sites into the plant genome has permitted the deletion, inversion, integration, and translocation of host and introduced DNA fragments. Recombinase-based strategies afford precise and predictable engineering of the plant genome

English Descriptors: Genetic engineering; DNA; Recombination process; Genetic transformation; Inversion; Deletion; Translocation; Gene rearrangement; Review

French Descriptors: Genie genetique; DNA; Processus recombinaison;